FRACTIONATION OF PEPSIN-DIGESTED DENATURED COLLAGEN

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The pepsin-digest of denatured rat-tail-tendon collagen gives seven well defined bands (A-G and α') in analytical starch gel electrophoresis (Penttinen, R. et al.: Acta chem. scand. 20, 1304, 1966). The fragments have been separated in preparative scale for characterization.

The digest was first divided in five preliminary molecular-size classes by molecular sieving on Sephadex G-200. The highest-molecular-weight fraction was then recycled 3—4 times on Sephadex G-200 to obtain the fragment α'. The middle-molecular-weight fraction was divided into several peaks by using the carboxymethylcellulose (CMC) technique (Piez, K. A. et al.: Biochemistry 2, 58, 1963). The last peak emerging from the CMC-column contained the band D-material highly enriched, and it was further purified by preparative starch-gel electrophoresis (Hollmén, T. and Kulonen, E.: Anal. Biochem. 14, 455, 1966). These fragments α' and D have been characterized (see the following abstract, Pikkarainen, J. et al.).

The two small-molecular-weight fractions have been chromatographed on phosphocellulose (Bornstein, P. and Piez, K. A.: Biochemistry 5, 3460, 1966) and are obtained in high purity after rechromatography. Electrophoretically homogeneous band A-material can be obtained by preparative starch gel electrophoresis directly after gel filtration, but it chromatographs as two distinct peaks on CMC.